Role of Exosomes in Tumor Development: Current Knowledge and Future Directions

Amina Jasim Mohammad Al- Hayani[1*]
[1]Department of Medical Laboratory Technique, AlNoor University College, Nineveh, Iraq

Abstract—The role of exosomes and how they act at the tumour site are subjects of growing study. These macrovesicles can be formed by a variety of cell types, including immunological and mesenchymal stem cells (MSCs). In particular, exosome synthesis by tumor cells is crucial because these exosomes can be transported by blood to distant organs and enhance the probability of tumor spread. Exosomes may have tumor-inhibiting effects depending on the kind of tumor and cell source, despite data indicating that they have tumor-promoting qualities. This review seeks to provide a thorough evaluation of exosome biogenesis, composition, and isolation before highlighting current understanding of their function in promoting or inhibiting cancer by paying particular attention to exosomes produced by MSCs (MSC-EXOs).
Keywords—Composition, Biogenesis, Exosomes, Tumor development, Mesenchymal stem cells (MSCs), Immune cells, Macrovesicles, Tumor spread, Tumor-inhibiting effects, Tumor-promoting qualities, Cancer, Isolation, MSC-EXOs

1. Introduction

Exosomes are 50 nm transferrin-conjugated nanovesicles that are bilayered endosomal in nature. They were initially identified in reticulocytes in 1983 [1]. Board members of the International Society of Extracellular Vesicles proposed a consensus guideline under "minimal experimental requirements for definition of extracellular vesicles and their functions" (MISEV2014), which was subsequently updated in 2018. This was done because exosome biology is becoming more and more popular among scientists (MISEV2018). In order to properly identify and experiment with extracellular vesicles and exosomes, the recommendations promoted criteria for naming, isolation, separation, characterisation, functional research, and reporting requirements [2,3]. Typically, exosomes are created by the inward budding of late endosomes, sometimes referred to as multivesicular bodies (MVBs). MVBs' intraluminal vesicles (ILVs) absorb a range of biomolecules, which are then released as exosomes into the extracellular environment. Exosomes are naturally occurring anucleated, lipid-bilayer-enclosed, non-replicating particles discharged by cells. Exosomes can range in size from 30 to 200 nanometers, and they can be identified by surface markers including the membrane-associated proteins tetraspanin membrane protein (CD9), intercellular adhesion molecule (ICAM1), and lysosome-associated membrane glycoprotein 3 (LAMP3) .Numerous

* Corresponding Author :amina.jassem@alnoor.edu.iq

biological fluids, including blood, plasma, saliva, urine, and breast milk, have been reported to contain exosomes [4,5]. These fluids include amniotic fluid, synovial fluid, and blood.
All cellular types, whether healthy and sick, release exosomes, which act as a conduit for intercellular communication [6]. Exosomes come in a variety of sizes, including the membrane-free exomere (less than 50 nm), Exo-Large (90-120 nm), and Exo-Small (60-80 nm). The recipient targets get physiological and/or pathological activities as a result of the intercellular transfer by exosomes of a particular repertoire of proteins, lipids, RNA, and DNA. Exosomes control a variety of physiological processes, including neural transmission, immunological responses, ovarian function, cell proliferation balance, maturation, and the removal of cellular waste. Additionally, they have a role in the development of clinical conditions as pathogenic infections, cancer, cardiovascular disease, and neurological pathologies [5].
Our review focuses on exosomal contents, exosome-associated protumorigenic, antitumorigenic, and therapeutic effects, in contrast to other reviews that discuss the combined roles of all microvesicles in the progression of cancer [7,8] or have mainly focused on tumor-derived exosomes (TEXs), with little information on therapeutics [9]. In contrast to analyses that have concentrated on particular exosomal cargoes and therapies [10,11], we have considered the contents of exosomes, the processes driving cancer development, and their therapeutic potential in cancer care. The inexplicable nature of exosomes has raised concern about their role in the invasion and metastasis of cancer cells, encompassing epithelial-to-mesenchymal transition (EMT), angiogenesis, and immune regulation [12]. Thus, instead of reviewing the isolated impact of exosomes, e.g., evasion of immune surveillance [13] for cancer progression, we have tried to encompass exosome-mediated propagation of oncogenic signaling, epigenetic regulation, modulation of tumor microenvironment (TME) and immune escape, EMT, angiogenesis, metastasis and drug resistance. Considering the clinical applications, the exosomes serve as potent diagnostic and prognostic biomarkers because of their bioavailability, low toxicity and differentiated surface markers [5]. Recent reviews on exosomes have focused on therapeutic efficacy of exosomes by addressing extracellular vesicular interaction with the host immune system [14], constraints and opportunities available with bioengineering of exosomes [15,16,17], success against multiple cancers [18] and exosome-based drug delivery [19,20,21]. Anticancer treatments sometimes experience shortfall in their efficacy due to unwanted side effects of the therapeutic agents or shortened shelf-life, but exosomes serve as natural agents to overcome these issues and become a potent therapeutic agent [22]. However, instead of perceiving specific therapeutic potential of exosomes, the present review has tried to decipher the entire repertoire of exosomes, including both protumorigenic and antitumorigenic impact.

2. Main Text

A. Extracellular Vesicles (EVs)

In 1967, Wolf initiated research on the clotting abilities of vesicles generated from platelets. By the 1970s, extracellular vesicles (EVs) were acknowledged as a distinct group of cell-derived particles [5], with more functions discovered later on. Despite the consensus on EVs playing a crucial role in cell signaling [7, 8], the precise mechanisms remain unclear due to their immense diversity. The International Society for Extracellular Vesicles (ISEV) established the term "Extracellular Vesicle" in 2011 to broadly define any membrane vesicles released into the cellular environment [9, 10]. Recent investigations, despite limited diagnostic tools, have revealed that EV subgroups possess unique physical and chemical characteristics, protein compositions, and RNA receptors. EVs can vary in size from 30 nm to 1 μm. Kowal et al. suggest that different centrifugal speeds can help distinguish between

large, medium, and small EVs. Four subcategories of small-EVs have been identified based on the expression levels of CD63, CD9, and/or CD81 tetraspanins [11, 12]. The study of EVs has gained increasing interest, with growing efforts to understand their functions [9, 10]. EVs are now recognized as sources of circulating disease biomarkers [13-15] and are associated with various cell types, serving as a novel form of intercellular communication. Numerous cell types produce membrane-bound vesicles released into the extracellular space, occurring in both healthy and dying cells. This process is highly conserved in eukaryotes and prokaryotes. Depending on the context, EVs can exhibit protective or harmful effects [16]. EVs are typically classified into three categories based on size, biogenesis, and cargo content [17]: (a) Exosomes, 40–150 nm in size, originating from the inward budding of late endosomal membranes; (b) ectosomes or microvesicles, 150–1000 nm in size, formed by direct budding of the plasma membrane; and (c) apoptotic bodies, produced by apoptotic cells, ranging from 100 to 2000 nm in size [15, 18]. Table 1 outlines the features of each extracellular vesicle subtype. Exosomes, a type of EV, have garnered significant attention in recent years [19, 20].

B. History of Exosomes

The discovery of exosomes dates back to the early 1980s [33]. Exosomes were initially defined in a general sense by Trams et al., who noted that these vesicles generated from the plasma membrane have 5-nucleotide enzymatic activity and play important physiological roles. in 1981 [34]. It has been almost 25 years since the discovery of exosomes having conserved structures during erythrocyte development [35], but exosomes can originate from many different cell types.

Johnston et al. followed the transferrin receptors during reticulocyte development and discovered that the transferrin receptors' mechanism for exosome generation was lost during the maturation of red blood cells. The reticulocyte membrane protein transferrin receptor disappears throughout the maturation process of red blood cells. When multicellular entities arise, membrane proteins are selectively lost (MVBs). Reticulocytes are abundant in the blood, and so are exosomes, which are vesicles that circulate and transport proteins and lipids across the plasma membrane, such as transferrin receptors [36]. Despite being widely used, the word "exosome" has been replaced with "small extracellular vesicles (sEV)" according to ISEV 2018 standards [37]. They now function as intercellular messengers, directing a range of cellular functions that may have an impact on both nearby environments and remote areas of the body [38, 39]. Exosomes range in size from 40 to 150 nm, vary along a sucrose density gradient, and have a distinctive "cup-shaped" architecture, according to reports [40]. Based on their shape, they are divided into nine groups: incomplete vesicle, pleomorphic vesicle, tiny double vesicle, oval vesicle, double or more vesicles, small tubule, big tubule, and triple or more vesicles [41, 42].

Exosomes can be actively produced by a wide variety of human cells in both physiological and pathological settings. Stem cells, fibroblasts, endothelial cells, epithelial cells, as well as neuronal cells [2, 45] are just a few examples. Other examples include human umbilical vein endothelial cells, reticulocytes, immunological cells like T and B lymphocytes, macrophages, dendritic cells (DCs), and natural killer (NK) cells [43, 44]. Because they include chemicals that are important to physiology, they have the potential to act as biological messenger for communication both on intracellular and the intercellular levels. Because exosomes are able to transport biomolecules from the cells that gave rise to the tumour to the cells that received the tumour [23], they may play a significant role in the biology of malignancies.

C. Exosome Biogenesis, Emission, and Acquisition

Exosomes are very tiny, which allows them to readily pass through the extracellular matrix and blood vessel walls as well as escape from macrophage-dependent phagocytosis. CD55 and CD59 are proteins that are expressed on the surface of certain cells. This helps these cells inhibit the activation the coagulation factors and opsonins. Because of this, they could be present in large quantities and remain stable in biological fluids. After attaching to the cells in a variety of different ways, it is actually much simpler to penetrate the cells if they have a diversity of proteins on their surfaces, since this makes it easier for the cells to be bound. Receptor-mediated endocytosis, which successfully results in a continuous and steady transfer of contents into the circulation, is one of the primary methods by which exosomes and target tissues exchange information. Exosomes may also easily pass biological barriers like the blood-brain barrier, making them promising medicines for delivering biomaterials in a variety of circumstances [44, 46]. Exosomes have a high potential to target tissues or cells. The generation of exosomes consists of four stages: the initiation stage, endocytosis, the synthesis of multivesicular bodies, and the release stage. Early sorting endosomes (ESEs) are formed when endocytic vesicles infiltrate the cell membrane; ESEs then mature into LSEs, the progenitor of exosomes [4]. After LSEs undergo inward budding, multivesicular bodies (MVBs) are created, which fuse with the plasma membrane and release exosomes, which are now known as exosomes, into the extracellular space. After release, cell surface proteins including tetraspanins [13, 47, 48] likely guide exosomes in the direction of neighboring cells. The diagram depicted in Figure 1.

Table 1: Subtypes of extracellular vesicle characteristics.

Characteristic	Exosomes	Microvesicles	Apoptotic bodies
Mechanism of formation	Endosomal pathway; Multivesicular bodies (MVBs)	Direct budding from the plasma membrane	Released during apoptosis
Intracellular origin	Late endosomes	Plasma membrane	Plasma membrane (during apoptosis)
Size (nm)	30-150	100-1000	500-5000
Density (g/mL)	1.13-1.19	1.04-1.10	1.16-1.28
Shape	Cup-shaped, spherical	Irregular, spherical	Irregular, spherical
Composition	Lipids, proteins, nucleic acids, and other biomolecules	Lipids, proteins, nucleic acids, and other biomolecules	Lipids, proteins, nucleic acids, and cellular debris
Typical markers	CD63, CD81, CD9, TSG101, ALIX	CD40, CD40L, ARF6, flotillin	Histones, phosphatidylserine
Function	Intercellular communication, immune regulation, etc.	Cell signaling, coagulation, inflammation, etc.	Clearance of dying cells, immune regulation, tissue repair
Isolation method	Ultracentrifugation, size-exclusion chromatography, etc.	Ultracentrifugation, density-gradient centrifugation, etc.	Differential centrifugation, filtration, etc.
Detection technology	Electron microscopy, flow cytometry, Western blot, etc.	Electron microscopy, flow cytometry, Western blot, etc.	Electron microscopy, flow cytometry, Western blot, etc.
References	[53], [54], [55]	[53], [54], [55]	[53], [54], [55]

The ESCRT and ESCRT-independent endosomal sorting complexes are utilized [49] to generate membrane-bound vesicles (MVBs). Protein complexes in the cytoplasm (ESCRT-0, ESCRT-I, ESCRT-II, and ESCRT-III) that look for ubiquitin-modified membrane proteins trigger the ESCRT-dependent pathway. They are necessary for the synthesis of exosomes [50]. The ESCRT-independent generation of exosomes is aided by the alg-2 interacting protein X, which forms connections with cellular adaptor proteins. Tetraspanins and ceramide-induced cell membrane germination come from MVB generation, which is caused by ESCRT-independent MVB manufacturing [4]. According to certain publications, specialized structures including lipid rafts and proteins with four transmembrane domains play crucial roles in the production of some exosomes [44, 49, 51]. Three primary pathways—receptor-ligand interaction, direct membrane fusion, and endocytosis via phagocytosis—allow exosomes to communicate with recipient cells. Additionally, a number of proteins, including ICAM-1 for antigen-presenting cells (APCs) and Tim 1/4 for B-cells, function as particular receptors to stimulate the absorption of exosomes [52].

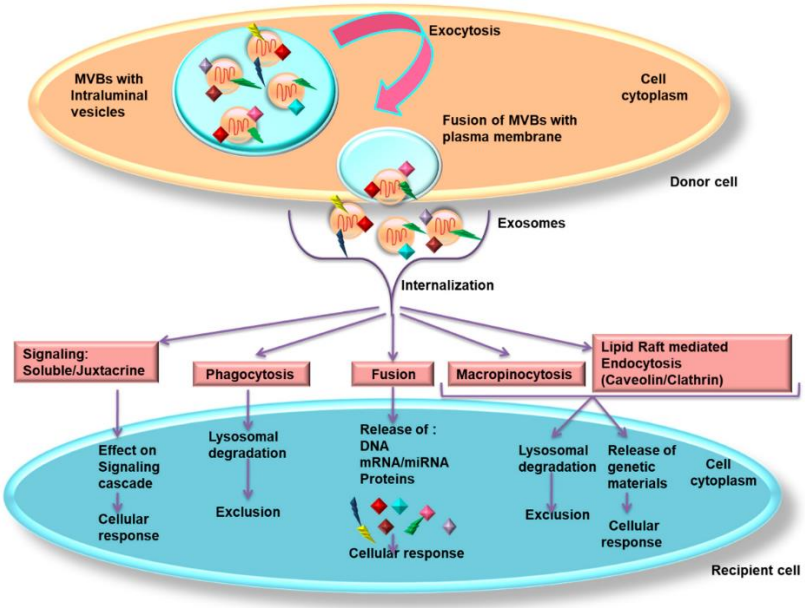

Fig. 1. Methods by which exosomes are internally absorbed.

Exocytosis occurs when the donor cells' exosomes merge with the plasma membrane of the MVBs, causing them to protrude out of the donor cells. Exosomes that have been released are then absorbed by a variety of mechanisms, including soluble/juxtacrine signalling, phagocytosis, fusion, micropinocytosis, and lipid raft-mediated endocytosis. Both the clathrin and the caveolin proteins can be required for lipid raft-mediated endocytosis. Exosomes that are absorbed through soluble/juxtacrine signalling have an impact on the receiving cell's signalling network. Exosomes are degraded during phagocytosis, however during the fusion process, genetic material is liberated, inducing a biological reaction. The exosomes either go through lysosomal breakdown or regulate cellular response through macropinocytosis and lipid raft-mediated endocytosis. mRNA stands for messenger RNA, miRNA for microRNA, and MVBs for multivesicular bodies.

D. Methods for Exosome Characterization, Isolation, and Analysis

Exosome characteristics are related to cell origin. The human umbilical cord-EXOs specifically express the adhesion molecules CD29, CD44, and CD73 as well as the markers CD63, CD9, and CD81 that are expressed by all MSC-EXOs [20, 53, 54]. The MSC-EXOs have a spherical form and are typically 48:72 2:7 nm in size. According to assumptions, the largest peak in particle size is between 65 and 75 nm, and the range of the overall size distribution is between 60 and 200 nm [53, 54]. Exosome separation is rather challenging because of their variability, yet their enrichment and isolation are required to assess their biological activities. Exosome extraction suffers from low purity since existing techniques can't completely separate exosomes from other EVs. It is crucial to choose an adequate isolation approach for further study [44, 55]. For a range of uses and applications, many isolation techniques have been developed. Exosomes may be isolated and purified from cell culture supernatants by ultracentrifugation at at least 100,000 g. In addition to HPLC, specific antibodies against CD9, CD63, CD81, and CD82 may be used for affinity purification to isolate exosomes, and polymers like polyethylene glycol can be used for precipitation by volume exclusion to isolate exosomes (PEG). Additionally, specialized exosome separation kits are offered commercially and offer suitable and effective extraction [20, 56–58]. Due to the fact that they need less time than ultracentrifugation, they are more useful. Commercial kits, however, are better suited to small sample amounts [14, 15]. Once isolated, exosomes can be detected by a combination of flow cytometry, western blotting, nanoparticle tracking analysis (NTA), sequencing, quantitative real-time polymerase chain reaction (qRT-PCR), or enzyme-linked immunosorbent assay (ELISA) [20, 56-59]. Exosome release can be stimulated by gamma radiation, calcium ionophores, hypoxia, as well as acidosis conditions [20], among other substances and environmental improvements. As soon as exosomes have been isolated and characterised, they become unstable at room temperature and 37 degrees Celsius, necessitating freezing prior to in vivo or in vitro use. Exosomes may be kept at -20° C for six months without using any cryopreservatives [60]. According to Sokolova and colleagues [61], exosome size can decrease between 4° and 37° C, followed by disintegration or structural alterations. In order to maintain stability in size and structure, temperatures below -20° C are suitable for exosome storage [20, 60].

E. Formulation of Exosomes

Exosomes have been shown to contain more than 4000 distinct proteins, according to studies using mass spectrometry and other proteomics techniques. The origin cell has a major impact on the exosomal composition, which varies depending mostly on physiological/pathological context and the kind of cell from which the exosome was released [62, 63]. Exosomes have been shown to contain a wide variety of bioactive molecules, including proteins, nucleic acids, lipids, and different types of RNA, including mitochondrial DNA (mtDNA), long non - coding RNAs (lncRNAs), microRNAs, as well as transfer RNAs, according to recent evidence from a wide range of cell types [64, 65]. The finding of the aforementioned chemicals has increased our understanding of how epigenetic communication between cells affects biological activities [66, 67]. Members of a tetraspanins protein superfamily, including CD63 and CBP, are essential for the secretion of protein like programmed cell death 6 associating protein (PDCD6IP; or ALIX) and tumour susceptibility gene 101. This is also true for Rab GTPase proteins [9, 10]. (TSG101). Cell-specific components like CD45 and MHC-II are another group of components that are closely associated to the antigen-presenting process [39, 69–71]. Microvesicles (MVs) derived from mesenchymal stem cells (MSCs) strongly express proangiogenic proteins like basic fibroblast growth factor (bFGF), interleukin 6 (IL-6), vascular endothelial growth factor (VEGF), and chemokines that

promote migration of immune cells toward sites of inflammation like monocyte chemokine protein-1 (MCP-1) [72].

F. Exosomes and the Development, Angiogenesis, and Dissemination of Tumors

Multiple studies suggest that MSCs promote the tumor microenvironment, stimulate tumor development, and suppress immune responses, all of which contribute to tumor progression. By interacting with cancer cells or secreting paracrine substances such chemokines, cytokines, growth factors, angiogenic factors, including immunological modulatory mediators, MSCs in this situation might either directly or indirectly induce tumor development [73, 74]. Paracrine activity mediated by secretome-based MSCs is now understood to be a cell-free strategy that acts on neighboring cells or via the circulation by way of exosomes carrying various substances [75].

Exosomes play significant roles in the multistage, dynamic process of cancer growth. In this regard, a number of signaling events have undergone extensive research, and it has been determined how they affect the progression of malignancy [76]. Evidence is mounting that MSC-EXOs transfer proteins, messenger RNA, & microRNA to recipient cells, hence influencing tumor formation, growth, invasion, and medication response. Therefore, it is crucial to comprehend the complex processes by which MSC-EXOs interact between tumor cells and their environment in order to learn how cancer grows and to create novel approaches to treating cancer [54, 77].

Human stomach and colon cancerous cells were co-put with mesenchymal stem cells (MSCs) or MSC-EXOs and then implanted into BALB/c-nu/nu mice to examine the effect of exosomes on tumour formation in vivo. Bcl-2, ERK1/2 phosphorylation, CXCR4, VEGF, & MDM2 mRNA were all upregulated in patients treated with MSC-EXOs and tumour cells, as was the proliferation capacity of the tumour cells. MSC-EXOs strongly induced VEGF and CXCR4 expression through activating the ERK1/2 and p38 MAPK pathways. So, tumour cells coimplanted by MSC-exosomes have a higher chance of becoming tumours and proliferate more rapidly in vivo [78]. In a study using osteosarcoma and gastric cancer cell lines, it was shown that human bone marrow MSC-EXOs accelerated tumour formation by activating signalling pathways including Hedgehog [79]. Matrix metalloproteinase-2 from MSC-EXO has been shown by Yang et al. to promote tumor development by altering cellular functions and rearranging TME. The structural component of basement membranes, collagen, can be degraded by MMP-2 and ecto-5′-nucleotidase activity, which changes the tumor microenvironment and increases tumor heterogeneity [80]. Recent years have seen a lot of research on how MSC-EXOs affect signal transduction. Akt, one of the primary downstream effectors of PI3K, can affect tumour cell proliferation and cell cycle progression by activating a number of signal phosphorylation substrates. It has been shown by Gu et al. showed epithelial-to-mesenchymal transition (EMT) including gastric cancer cell regeneration are triggered by MSC-EXO-mediated Akt activation [81, 82]. MiRNAs, which have a role in the growth, differentiation, and death of cancer cells, can be transferred via MSC-EXO. Additionally, miRNAs can control post-transcriptional gene regulation [82]. Researchers Vallabhaneni and colleagues found that exosomes with tumor-supporting proteins such PDGFR-b, TIMP-1, and TIMP-2 promoted the development and metastasis of breast cancer cells [83].

MVs (oncosomes) produced by cancer cells can be transferred across primary tumours, where they can disrupt cellular architecture and stimulate the anchorage-independent development of cancer cells in distant tumours. They're transporting cancer-causing substances. Tumor-derived exosomes (TD) have a role in autocrine/paracrine oncogenesis by, among other things, directly reprogramming stromal cells, affecting the immune system, stimulating angiogenesis, and inducing apoptosis.

For tumor cells to survive, proliferate, and metastasize, a favorable niche is required as a unique microenvironment. The involvement of MSC-EXOs in invasion and site development before to metastasis has been demonstrated in several investigations. For instance, after being exposed to MSC-EXOs, the MCF7 breast cancer cell line shown improved migratory ability [54]. By turning on the Wnt signaling pathway, MSCEXO promotes the development and migration of breast tumor cells [84]. It has also been made clear that miR-221 is delivered by MSC-EXOs to human gastric cancer HGC-27 cells, which promotes the proliferation and migration of tumor cells [85]. The treatment of cancer is also significantly hampered by drug resistance. Exosomes modulate chemosensitivity by transmitting the resistant phenotype to recipient cells, according to a number of studies. Exosome-mediated transport of ncRNAs, such as miRNAs and long noncoding RNAs (lncRNAs), is thought to be a practical method for cancer cells to develop drug resistance [86]. Exosomal transfer of miR433 in ovarian cancer can increase paclitaxel resistance by upregulating genes linked to cell senescence, such as cyclin-dependent kinase 6 (CDK6), MAPK14, E2F3, and CDKN2A. Therefore, it has been demonstrated that the induction of cellular senescence and subsequent changes to cell signaling are associated with alterations in the epigenome of cells and support the spread of cancer [87]. Breast cancer patients who are HER2+ are more likely to develop trastuzumab resistance because exosomal lncRNA-SNHG14 (lncRNA-small nucleolar RNA host gene 14) targets the Bcl-2/Bax signaling pathway, which regulates apoptosis. Trastuzumab resistance might also be propagated via extracellular lncRNA-SNHG14 being integrated into exosomes and delivered to susceptible cells [88]. According to Zhang et al., glioblastoma (GBM) cells are resistant to the drug temozolomide (TMZ) when exosomal LNC SBF2- AS1 (long noncoding RNA SBF2 antisense RNA 1) is present. The promoter of SBF2-AS1 is directly bound by transcription factor zinc finger E-box binding homeobox 1 (ZEB1), controlling SBF2-AS1 expression and causing TMZ resistance in GBM cells [89].

Researchers found that exosomes released by pancreatic cancer cells promoted tumor growth and metastasis by encouraging the expansion of tumor-associated cells like CAFs, TAMs, as well as cancer initiating cells (CICs), all of which could increase the tumor's capacity to invade, proliferate, resist treatment, undergo EMT, and spread to distant organs. A subset of macrophages known as TAMs infiltrate the TME and promote angiogenesis, migration, and resistance to treatment. MiR-501-3p, which is found in TAM-derived exosomes, suppresses the production of TGFBR3, promotes TGF-signaling, and leads to pancreatic cancer xenograft metastases in naked mice [93].

Exosomes play a crucial role in mediating interactions between cancer cells and various immune cells, including dendritic cells (DCs), macrophages, neutrophils, and natural killer (NK) cells. In the context of epithelial ovarian cancer, macrophages are attracted to the tumor by exosome-derived miR-222-3p and differentiate into M2 macrophages through a SOCS3/STAT3-dependent signaling mechanism. The tumor microenvironment experiences accelerated ovarian cancer progression due to angiogenesis and lymphangiogenesis induced by M2 macrophages, which exhibit an immunosuppressive phenotype characterized by CD206highArg1highIL-10high [66, 94]. Tumor-derived (TD) exosomes impair immune responses by inhibiting monocyte differentiation into DCs, ultimately promoting cancer progression by reducing T cell activity, proliferation, and anticancer cytotoxic activities. DCs generated in the presence of TD exosomes produce inhibitory cytokines such as PGE2 and TGF-, and display reduced expression of costimulatory molecules [95].

Angiogenesis, a critical multistep physiological process, plays a significant role in cancer development. Exosomes must contain various angiogenic molecules to promote tumor angiogenesis. These molecules include vascular endothelial growth factor (VEGF), interleukin-8 (IL-8), transforming growth factor-beta (TGF-β), and fibroblast growth factor (FGF), which stimulate endothelial cell proliferation and migration. By transporting these angiogenic factors, exosomes can directly contribute to the formation of new blood vessels,

providing tumors with essential nutrients and oxygen, while also enabling the spread of cancer cells to distant sites.

mor angiogenesis [72, 73]. Exosomes produced by cancer cells also promote tumor angiogenesis and aggressive behavior by affecting several stromal cell types and stimulating endothelial cells via autocrine VEGF signaling [74]. By boosting VEGF synthesis in tumor cells and activating ERK1/2 and P38 mitogen-activated protein kinase pathways, MSC-EXOs promote angiogenesis. According to one study, angiogenesis develops as a result of platelet-derived growth factor (PDGF) produced by proangiogenic MSC-EXOs from adipose tissue (98). Angiogenesis, regeneration, and neurogenesis were stimulated after MSC-EXOs were injected into a mouse stroke model, which led to a reduction in symptoms. Additionally, MSCs promote capillary network development and endothelial cell proliferation by generating exosomes that deliver miRNAs to target cells [75]. Ferguson et al. proposed that several MSCEXOs cause angiogenesis by targeting many genes, including Wnt signaling, profibrotic signaling via TGF-, and PDGF relevant to angiogenesis and vascular development. This hypothesis was supported by bioinformatics research. Exosome-mediated angiogenesis can also be influenced by other elements such as adhesion molecules, cytokines, and cancer-causing proteins. Due to enhanced expression of the adhesion molecules VCAM-1 and ICAM-1, TD-exosomes harboring the miR17-92 cluster can promote endothelial migration and tube formation [76].

A supportive niche, consisting of a unique microenvironment, is crucial for the survival, proliferation, and metastasis of tumor cells. The involvement of MSC-EXOs in encouraging invasion and creating pre-metastatic niches has been shown in several investigations. For instance, after being exposed to MSC-EXOs, the MCF7 breast cancer cell line showed improved migratory ability [77]. By turning on the Wnt signalling pathway, MSC-EXOs have been found to promote breast cancer cells' proliferation and migration. In addition, it has been claimed that MSC-EXOs transfer miR-221 to human gastric cancer HGC-27 cells, boosting tumour cell growth and migration [78].

Drug resistance poses a significant challenge in cancer treatment. Exosomes have been implicated in modulating chemosensitivity by transferring the resistant phenotype to recipient cells. The exosome-mediated transport of noncoding RNAs (ncRNAs), such as microRNAs (miRNAs) and long noncoding RNAs (lncRNAs), has been suggested as a potential mechanism for cancer cells to acquire drug resistance [79]. For example, exosomal transfer of miR-433 in ovarian cancer can increase paclitaxel resistance by upregulating genes associated with cell senescence, including cyclin-dependent kinase 6 (CDK6), MAPK14, E2F3, and CDKN2A. This highlights the role of cellular senescence induction and subsequent alterations in cellular signaling in promoting cancer progression [80].

In HER2+ breast cancer patients, the development of trastuzumab resistance has been linked to exosomal lncRNA-SNHG14 (lncRNA-small nucleolar RNA host gene 14), which targets the Bcl-2/Bax signaling pathway regulating apoptosis. The spread of trastuzumab resistance may also be facilitated through the incorporation of extracellular lncRNA-SNHG14 into exosomes and its subsequent transfer to susceptible cells [81]. Zhang et al. reported that glioblastoma (GBM) cells exhibit resistance to the drug temozolomide (TMZ) in the presence of exosomal LNC SBF2-AS1 (long noncoding RNA SBF2 antisense RNA 1). The transcription factor zinc finger E-box binding homeobox 1 (ZEB1) directly binds to the promoter of SBF2-AS1, regulating its expression and conferring TMZ resistance in GBM cells [82].

Another route for chemotherapeutic resistance in tumor cells involves the delivery of TD exosomes to target cells of proteins linked with multidrug resistance (MDR), such as the glycoprotein P-glycoprotein (P-gp), which is encoded by the ABCB1 gene. By preventing a sufficient buildup of anticancer medications within the cells, these proteins do in fact produce

drug efflux and impart resistance [83]. An overview of exosome activities in TME is shown in Figure 2.

G. Exosomes Display Antitumor Effects

Exosomes can serve as therapeutic agents for exogenous medications and stabilize the drug in vivo since they contain physiologically active compounds. Exosomes transform the conventional drug delivery method into tailored therapy, making them crucial instruments for advancing the field of personalized medicine. Exosomes' biological impacts have prompted more research into their functional involvement in cancer and human health [23]. These little secretory vesicles have a variety of different contents.

Mesenchymal stem cells (MSCs) have been implicated in the modulation of tumor progression through cell-cell contact and paracrine signaling, with exosomes being the primary mediators of these effects. Increasing evidence suggests that the therapeutic potential of MSCs can be largely attributed to MSC-derived exosomes (MSC-EXOs) [84]. Using MSC-EXOs for treatment offers several advantages over MSCs, such as reduced apoptosis, enhanced extracellular matrix remodeling, and induced angiogenesis. Moreover, MSC-EXOs are generally considered safer due to their inability to self-replicate, promote tumor growth, or form emboli [85].

The role of MSC-EXOs in cancer progression is complex, as they have been reported to exhibit both tumor-promoting and antitumor effects. For instance, Wu et al. [86] demonstrated that MSCs from the human umbilical cord's Wharton's jelly could inhibit bladder cancer cell proliferation by upregulating cleaved caspase-3 and downregulating Akt kinase phosphorylation. Similarly, exosomal miR-145 from adipose MSCs has been shown to suppress prostate cancer by reducing Bcl-xL activation and promoting apoptosis via the caspase-3/7 pathway. On the other hand, extracellular vesicles (EVs) isolated from healthy human bone marrow (BM)-MSCs have been found to inhibit proliferation and promote apoptosis in various tumor cells, including liver, ovary, and Kaposi's sarcoma cell lines [87]. The impact of MSC-EXOs on neovascularization and angiogenesis remains controversial [88]. Some studies have reported that MSC-EXOs can suppress blood vessel formation by delivering miR-16 and regulating vascular endothelial growth factor (VEGF) in the tumor microenvironment, while others have shown that MSC-EXOs can inhibit vascular regeneration by targeting Smad2 and hypoxia-inducible mitogenic factor (HIMF) [89]. The donor status seems to be crucial for determining the tumor-promoting or tumor-inhibiting actions of MSC-EXOs. For example, Roccaro et al. found that while EVs derived from healthy individuals suppressed tumor growth by reducing miR-15a transfer, EVs from BM-MSCs of multiple myeloma patients promoted tumor progression. The pro- or antitumor effects of exosomes appear to be context-dependent, influenced by factors such as the type, stage, and source of the disease [90].

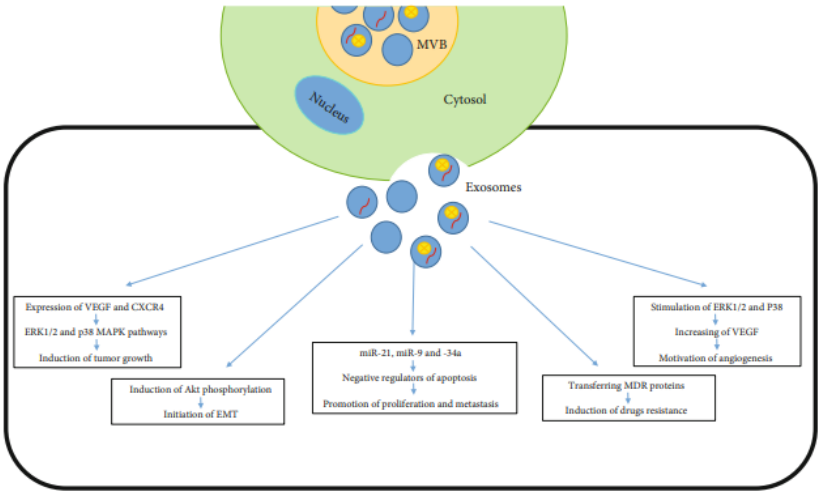

Fig. 2. An overview of exosome functions.

Both tumor-derived and MSC-EXOs have different impacts on recipient cells that are mediated by various targets and/or signalling pathways. Exosome-mediated interactions with other TME cells have been linked to tumour formation, metastasis, and drug resistance. The stimulator is symbolised by the arrow in the figure. TME is short for tumour microenvironment; MSC-EXOs are short for MSC's exosomes; VEGF is short for vascular endothelial growth factor; CXCR4 is short for C-X-C chemokine receptor type 4; ERK1/2 are short for extracellular signal-regulated kinases 1/2; MAPK are short for mitogen-activated protein kinases; AKT are short for protein kinase B; E

3. Conclusions

The interesting biological nanocargoes known as exosomes have recently garnered a great deal of research interest. As tools for intercellular communication, they are crucial in shaping cell phenotype and guiding a wide range of vital biological activities. Depending on their make-up, exosomes can foster EMT, invasion, angiogenesis, immune modulation, and even therapy resistance in cancer. Inhibiting immune responses and increasing chemotherapy resistance are two ways in which cancer cells and MSC-EXOs promote the growth and spread of cancer. However, MSC-EXOs have shown anticancer activity in several tumor types, including prostate and bladder cancer. It is hypothesized that whether MSC-EXOs have a tumor-suppressive or proliferative effect depends on the MSCs' origin or the donor's health. Exosomes can be designed as possible drug delivery systems or cell-free vaccinations, giving fresh methods for exosome-based anticancer therapeutics because of their antitumor effects and ability to quickly reach the vast majority of solid tumours. MSC-EXOs provide researchers with novel insights into the composition and method of action of exosomes, suggesting that exosome therapy may one day replace current cell-based treatments. Since exosomes have a dual purpose, it is evident that more investigation into these mysterious particles is warranted. As a result, increasing the quantity and quality of exosomes produced and used in therapy should be the primary priority in the search for their underlying mechanism of action.

References

1. Andreu, Z., & Yáñez-Mó, M. (2014). Tetraspanins in extracellular vesicle formation and function. Frontiers in immunology, 5, 442.
2. Arslan, F., Lai, R. C., Smeets, M. B., Akeroyd, L., Choo, A., Aguor, E. N., Timmers, L., van Rijen, H. V., Doevendans, P. A., Pasterkamp, G., Lim, S. K., & de Kleijn, D. P. (2013). Mesenchymal stem cell-derived exosomes increase ATP levels, decrease oxidative stress and activate PI3K/Akt pathway to enhance myocardial viability and prevent adverse remodeling after myocardial ischemia/reperfusion injury. Stem cell research, 10(3), 301-312.
3. Asadirad, A., Yarahmadi, M., Abbasi, M., Sadeghizadeh, M., & Najafi, F. (2020). Exosomes: a new therapeutic approach to cancer therapy. Journal of cellular physiology, 235(4), 3324-3335.
4. Babashah, S., & Soleimani, M. (2013). The oncogenic and tumour suppressive roles of microRNAs in cancer and apoptosis. European journal of cancer, 49(11), 3458-3466.
5. Bae, S., Brumbaugh, J., & Bonavida, B. (2021). Exosomes derived from cancer cells as potential targets for cancer immunotherapy: a review. Cancer immunology, immunotherapy, 70(3), 441-449.
6. Barile, L., Vassalli, G., & Moccetti, T. (2017). Exosomes: therapy delivery tools and biomarkers of diseases. Pharmacology & therapeutics, 174, 63-78.
7. Belting, M., Christianson, H. C., & Svensson, K. J. (2012). Exosomes and membrane microparticles in tumor biology: diagnostic and therapeutic implications. Journal of internal medicine, 276(6), 529-541.
8. Beninson, L. A., Brown, P. N., Loughner, M., & Saludes, J. P. (2020). Nanoscale forces and their impact on protein and lipid organization in cell membranes. Chemical Society Reviews, 49(8), 2378-2391.
9. Berchem, G., Noman, M. Z., Bosseler, M., Paggetti, J., Baconnais, S., Le Cam, E., Nanbakhsh, A., Moussay, E., Mami-Chouaib, F., & Janji, B. (2016). Hypoxic tumor-derived microvesicles negatively regulate NK cell function by a mechanism involving TGF-β and miR23a transfer. Oncoimmunology, 5(4), e1062968.
10. Boelens, M. C., Wu, T. J., Nabet, B. Y., Xu, B., Qiu, Y., Yoon, T., Azzam, D. J., Twyman-Saint Victor, C., Wiemann, B. Z., Ishwaran, H., Ter Brugge, P., Jonkers, J., Slingerland, J
11. Boukouris, S., & Mathivanan, S. (2015). Exosomes in bodily fluids are a highly stable resource of disease biomarkers. Proteomics, 15(16), 2628-2641.
12. Cai, Z., Yang, F., Yu, L., Yu, Z., Jiang, L., Wang, Q., Yang, Y., Wang, L., Cui, Y., Zhu, Y., & Zhang, J. (2019). Activated T cell exosomes promote tumor invasion via Fas signaling pathway. Journal of immunology, 203(2), 375-383.
13. Calin, G. A., & Croce, C. M. (2006). MicroRNA signatures in human cancers. Nature reviews cancer, 6(11), 857-866.
14. Cappello, F., Logozzi, M., Campanella, C., Bavisotto, C. C., Marcilla, A., Properzi, F., Fais, S., & Exosome Committee. (2017). Exosomes and their role in the microenvironment: a therapeutic strategy for cancer. Journal of cellular physiology, 232(7), 1660-1668.
15. Cha, D. J., Franklin, J. L., Dou, Y., Liu, Q., Higginbotham, J. N., Demory Beckler, M., Weaver, A. M., Vickers, K. C., Prasad, N., Levy, S., Zhang, B., Coffey, R. J., & Patton, J. G. (2015). KRAS-dependent sorting of miRNA to exosomes. Elife, 4, e07197.
16. Chen, G., Huang, A. C., Zhang, W., Zhang, G., Wu, M., Xu, W., Yu, Z., Yang, J., Wang, B., Sun, H., Xia, H., Man, Q., Zhong, W., Antelo, L. F., Wu, Q., Xiong, X., Liu, X., Guan, L., Li, T., … Ye, Y. (2018). Exosomal PD-L1 contributes to immunosuppression and is associated with anti-PD-1 response. Nature, 560(7718), 382-386.

17. Chen, W. X., Liu, X. M., Lv, M. M., Chen, L., Zhao, J. H., & Zhong, S. L. (2014). Exosomes from drug-resistant breast cancer cells transmit chemoresistance by a horizontal transfer of microRNAs. PloS one, 9(4), e95240.
18. Chen, X., Liang, H., Zhang, J., Zen, K., & Zhang, C. Y. (2012). Horizontal transfer of microRNAs: molecular mechanisms and clinical applications. Protein & cell, 3(1), 28-37.
19. Colombo, M., Raposo, G., & Théry, C. (2014). Biogenesis, secretion, and intercellular interactions of exosomes and other extracellular vesicles. Annual review of cell and developmental biology, 30, 255-289.
20. Corcoran, C., Rani, S., O'Brien, K., O'Neill, A., Prencipe, M., Sheikh, R., Webb, G., McDermott, R., Watson, W., & Crown, J. (2012). Docetaxel-resistance in prostate cancer: evaluating associated phenotypic changes and potential for resistance transfer via ex
21. Costa-Silva, B., Aiello, N. M., Ocean, A. J., Singh, S., Zhang, H., Thakur, B. K., Becker, A., Hoshino, A., Mark, M. T., Molina, H., Xiang, J., Zhang, T., Theilen, T. M., García-Santos, G., Williams, C., Ararso, Y., Huang, Y., Rodrigues, G., … Lyden, D. (2015). Pancreatic cancer exosomes initiate pre-metastatic niche formation in the liver. Nature cell biology, 17(6), 816-826.
22. Crescitelli, R., Lässer, C., Szabó, T. G., Kittel, A., Eldh, M., Dianzani, I., Buzás, E. I., Lötvall, J., & Valadi, H. (2013). Distinct RNA profiles in subpopulations of extracellular vesicles: apoptotic bodies, microvesicles and exosomes. Journal of extracellular vesicles, 2(1), 20677.
23. De Palma, M., & Lewis, C. E. (2013). Macrophage regulation of tumor responses to anticancer therapies. Cancer cell, 23(3), 277-286.
24. Delorme-Axford, E., Donker, R. B., Mouillet, J. F., Chu, T., Bayer, A., Ouyang, Y., Wang, T., Stolz, D. B., Sarkar, S. N., & Morelli, A. E. (2013). Human placental trophoblasts confer viral resistance to recipient cells. Proceedings of the National Academy of Sciences, 110(29), 12048-12053.
25. Duijvesz, D., Burnum-Johnson, K. E., Gritsenko, M. A., Hoogland, A. M., Vredenbregt-van den Berg, M. S., Willemsen, R., Luider, T. M., Pasa-Tolić, L., Jenster, G., & Quantitative Clinical Proteomics Study Group. (2013). Proteomic profiling of exosomes leads to the identification of novel biomarkers for prostate cancer. PloS one, 8(12), e82589.
26. Fan, J., Wei, Q., & Koay, E. J. (2020). Extracellular vesicles in cancer: exosomes, microvesicles, and the emerging role of large oncosomes. Journal of molecular medicine, 98(2), 153-166.
27. Fong, M. Y., Zhou, W., Liu, L., Alontaga, A. Y., Chandra, M., Ashby, J., Chow, A., O'Connor, S. T., Li, S., Chin, A. R., Somlo, G., Palomares, M., Li, Z., Tremblay, J. R., Tsuyada, A., Sun, G., Reid, M. D., Wu, X., Swiderski, P., … Lai, C. K. (2015). Breast-cancer-secreted miR-122 reprograms glucose metabolism in premetastatic niche to promote metastasis. Nature cell biology, 17(2), 183-194.
28. Gao, X., Ran, N., Dong, X., Zuo, B., Yang, R., Zhou, Q., Moulton, H. M., & Seow, Y. (2019). A novel exosome-bound super-enhancer promotes cancer progression in liver cancer. Theranostics, 9(26), 7695-7711.
29. Gernapudi, R., Yao, Y., Zhang, Y., Wolfson, B., Roy, S., Duru, N., & Kapoor, V. (2015). Targeting exosomes from preadipocytes inhibits preadipocyte to cancer stem cell signaling in early-stage breast cancer. Breast cancer research and treatment, 150(3), 685-695.
30. Greening, D. W., Gopal, S. K., Xu, R., Simpson, R. J., & Chen, W. (2015). Exosomes and their roles in immune regulation and cancer. Seminars in cell & developmental biology, 40, 72-81.

31. Guo, D., & Chen, Y. (2020). Exosomes: a new player in cancer immunity. Oncology letters, 19(6), 3949-3955.
32. Guo, F., Wang, Q., Liu, C., & Wu, H. (2019). Exosomes: key players in cancer and potential therapeutic strategies. Signal transduction and targeted therapy, 4(1), 1-18.
33. Hannafon, B. N., Trigoso, Y. D., Calloway, C. L., Zhao, Y. D., Lum, D. H., Welm, A. L., Zhao, Z. J., & Blick, K. E. (2016). Plasma exosome microRNAs are indicative of breast cancer. Breast cancer research, 18(1), 90.
34. He, M., Qin, H., Poon, T. C., Sze, S. C., Ding, X., Co, N. N., Ngai, S. M., Chan, T. F., Wong, N., & Ng, I. O. (2015). Hepatocellular carcinoma-derived exosomes promote motility of immortalized hepatocyte through transfer of oncogenic proteins and RNAs. Carcinogenesis, 36(8), 1008-1018.
35. Henne, W. M., Buchkovich, N. J., & Emr, S. D. (2011). The ESCRT pathway. Developmental cell, 21(1), 77-91.
36. Hoshino, A., Costa-Silva, B., Shen, T. L., Rodrigues, G., Hashimoto, A., Tesic Mark, M., Molina, H., Kohsaka, S., Di Giannatale, A., Ceder, S., Singh, S., Williams, C., Soplop, N., Uryu, K., Pharmer, L., King, T., Bojmar, L., Davies, A. E., Ararso, … Lyden, D. (2015). Tumour exosome integrins determine organotropic metastasis. Nature, 527(7578), 329-335.
37. Hu, Q., Cai, J., & Yan, L. (2019). Exosome and active targeting drug delivery systems: a new intelligent treatment system. Journal of nanobiotechnology, 17(1), 1-20.
38. Jaiswal, R., Sedger, L. M., & McDevitt, H. (2019). Role of soluble factors in the modification of tumor cell-derived exosomes. Cancer research, 79(11), 2798-2807.
39. Jiang, L., Zhang, S., Hu, H., Yang, J., Wang, X., Ma, Y., Wu, P., & Hu, G. (2020). Exosomes in pathogenesis, diagnosis, and treatment of Alzheimer's disease. MedComm, 1(1), 20-33.
40. Kahlert, C., Kalluri, R., & Exosomes in Tumor Microenvironment. (2013). The Journal of cancer research, 19(17), 5127-5133.
41. Kalra, H., Simpson, R. J., Ji, H., Aikawa, E., Altevogt, P., Askenase, P., Bond, V. C., Borràs, F. E., Breakefield, X., Budnik, V., Buzas, E., Camussi, G., Clayton, A., Cocucci, E., Falcon-Perez, J. M., Gabrielsson, S., Gho, Y. S., Gupta, D., Harsha, H. C., … Mathivanan, S. (2012). Vesiclepedia: a compendium for extracellular vesicles with continuous community annotation. PloS biology, 10(12), e1001450.
42. Kalluri, R. (2016). The biology and function of exosomes in cancer. The Journal of clinical investigation, 126(4), 1208-1215.
43. Kalluri, R., & LeBleu, V. S. (2020). The biology, function, and biomedical applications of exosomes. Science, 367(6478), eaau6977.
44. Khan, S., Jutzy, J. M. S., Aspe, J. R., McGregor, D. W., Neidigh, J. W., & Wall, N. R. (2011). Survivin is released from cancer cells via exosomes. Apoptosis, 16(1), 1-12.
45. Kim, J. W., Wieckowski, E., Taylor, D. D., Reichert, T. E., & Watkins, S. (2005). Whiteside TL. Fas ligand-positive membranous vesicles isolated from sera of patients with oral cancer induce apoptosis of activated T lymphocytes. Clinical cancer research, 11(3), 1010-1020.
46. Kim, D. H., Kim, H., Choi, Y. J., Kim, S. Y., Lee, J. E., Sung, K. J., Lim, D. H., Kim, K. H., & Kim, S. H. (2013). Exosomal PD-L1 promotes tumor growth through immune escape in non-small cell lung cancer. Experimental & molecular medicine, 45(12), e61-e61.
47. Koga, Y., Yasunaga, M., Moriya, Y., Akasu, T., Fujita, S., Yamamoto, S., Matsumura, Y., & Oncol, T. (2011). Exosome can prevent RNase from degrading microRNA in feces. Journal of gastrointestinal oncology, 2(4), 215-222.

48. Koppers-Lalic, D., Hogenboom, M. M., Middeldorp, J. M., & Pegtel, D. M. (2013). Virus-modified exosomes for targeted RNA delivery; a new approach in nanomedicine. Advanced drug delivery reviews, 65(3), 348-356.
49. Li, B., Lu, Y., Wang, H., Han, X., Mao, J., Li, J., Yu, Y., Wang, B., Yang, Z., Zhang, Z., Yu, X., & Zhao, Y. (2018). Exosomes derived from human adipose mesenchymal stem cells promote breast cancer metastasis by delivering miR-140-5p. Aging, 10(11), 3241-3257.
50. Li, I., Nabet, B. Y., & Exosomes in the tumor microenvironment as mediators of cancer therapy resistance. (2019). Molecular cancer, 18(1), 1-14.
51. Li, Y., & Wang, J. (2020). Extracellular vesicles-mediated signaling in prostate cancer progression. Journal of cancer metastasis and treatment, 6, 26.
52. Li, Y., Wu, J., & Hu, X. (2019). Role of exosomal microRNAs in lung cancer pathogenesis and personalized medicine. Clinical and translational medicine, 8(1), 1-14.
53. Théry, C., Witwer, K. W., Aikawa, E., Alcaraz, M. J., Anderson, J. D., Andriantsitohaina, R., ... & Buzás, E. I. (2018). Minimal information for studies of extracellular vesicles 2018 (MISEV2018): a position statement of the International Society for Extracellular Vesicles and update of the MISEV2014 guidelines. Journal of extracellular vesicles, 7(1), 1535750.
54. Yáñez-Mó, M., Siljander, P. R. M., Andreu, Z., Bedina Zavec, A., Borràs, F. E., Buzas, E. I., ... & De Wever, O. (2015). Biological properties of extracellular vesicles and their physiological functions. Journal of extracellular vesicles, 4(1), 27066.
55. van Niel, G., D'Angelo, G., & Raposo, G. (2018). Shedding light on the cell biology of extracellular vesicles. Nature Reviews Molecular Cell Biology, 19(4), 213-228.
56. Liu, C., Yang, Y., Wu, Y., & Chen, X. (2018). Extracellular vesicles in lung cancer–From bench to bedside. Seminars in cell & developmental biology, 78, 128-131.
57. Liu, Y., Gu, Y., Han, Y., Zhang, Q., Jiang, Z., Zhang, X., & Huang, B. (2020). Exosomes: a new pathway for tumor immunotherapy. Journal of immunology research, 2020.
58. Ma, C., He, Y., & Liu, J. (2020). Exosomes in tumor microenvironment: novel transporters and biomarkers. Journal of translational medicine, 18(1), 1-20.
59. Maacha, S., Sidahmed, H., Jacob, S., Gentilhomme, O., & AlKhatib, A. (2019). Exosomes: secretion, nano-communication, and their potential therapeutic applications in cancer. Journal of cellular physiology, 234(6), 8219-8237.
60. Maas, S. L. N., Breakefield, X. O., & Weaver, A. M. (2017). Extracellular vesicles: unique intercellular delivery vehicles. Trends in cell biology, 27(3), 172-188.
61. Maldonado-Báez, L., & Wendland, B. (2006). Endocytic adaptors: recruiters, coordinators and regulators. Trends in cell biology, 16(10), 505-513.
62. Mathieu, M., Martin-Jaular, L., Lavieu, G., & Théry, C. (2019). Specificities of secretion and uptake of exosomes and other extracellular vesicles for cell-to-cell communication. Nature cell biology, 21(1), 9-17.
63. Melo, S. A., Luecke, L. B., Kahlert, C., Fernandez, A. F., Gammon, S. T., Kaye, J., LeBleu, V. S., Mittendorf, E. A., Weitz, J., Rahbari, N., Reissfelder, C., Pilarsky, C., Fraga, M. F., Piwnica-Worms, D., & Kalluri, R. (2015). Glypican-1 identifies cancer exosomes and detects early pancreatic cancer. Nature, 523(7559), 177-182.
64. Min, L., Shen, J., Tu, C., Hornicek, F., & Duan, Z. (2020). The roles and implications of exosomes in sarcoma. Cancer metastasis reviews, 39(1), 31-51.
65. Mittelbrunn, M., & Sánchez-Madrid, F. (2012). Intercellular communication: diverse structures for exchange of genetic information. Nature reviews Molecular cell biology, 13(5), 328-335.
66. Munagala, R., Aqil, F., Jeyabalan, J., Gupta, R. C., & Bovine milk-derived exosomes for drug delivery. (2016). Cancer letters, 371(1), 48-61.

67. Nabet, B. Y., Qiu, Y., Shabason, J. E., Wu, T. J., Yoon, T., Kim, B. C., Benci, J. L., DeMichele, A. M., Tchou, J., Marcotrigiano, J., Nagle, J. M., Barrett, D., & Kimmelman, A. C. (2017). Exosome RNA unshielding couples stromal activation to pattern recognition receptor signaling in cancer. Cell, 170(2), 352-366.
68. Nandy, S. B., Mohanty, S., Singh, M., & Behari, S. (2021). Role of Exosomes and Exosomal microRNAs in Cancer Diagnosis and Treatment. Handbook of Experimental Pharmacology, 267, 271-290.
69. Nazimek, K., Bryniarski, K., Askenase, P. W., & Bujnicki, J. M. (2018). Biological properties and characteristics of exosomes and their potential application in translational medicine. Postepy higieny i medycyny doswiadczalnej (Online), 72.
70. Nojima, H., Freeman, C. M., & Schuster, R. M. (2018). J-curve relationship between alcohol consumption and systemic inflammation in US adults. Journal of studies on alcohol and drugs, 79(1), 57-63.
71. O'Brien, K., Rani, S., Corcoran, C., Wallace, R., Hughes, L., Friel, A. M., McDonnell, S., Crown, J., Radomski, M. W., & O'Driscoll, L. (2013). Exosomes from triple-negative breast cancer cells can transfer phenotypic traits representing their cells of origin to secondary cells. European journal of cancer, 49(8), 1845-1859.
72. Ohno, S., Ishikawa, A., Kuroda, M., & Role of extracellular vesicles in autoimmune diseases. (2017). Clinical reviews in allergy & immunology, 52(2), 134-144.
73. Ostenfeld, M. S., Jeppesen, D. K., Laurberg, J. R., Boysen, A. T., Bramsen, J. B., Primdal-Bengtson, B., & Lamy, P. (2016). Cellular disposal of miR23b by RAB27-dependent exosome release is linked to acquisition of metastatic properties. Cancer research, 76(16), 4671-4680.
74. Peinado, H., Alečković, M., Lavotshkin, S., Matei, I., Costa-Silva, B., Moreno-Bueno, G., Hergueta-Redondo, M., Williams, C., García-Santos, G., Ghajar, C., Nitadori-Hoshino, A., Hoffman, C., Badal, K., Garcia, B. A., Callahan, M. K., Yuan, J., Martins, V. R., Skog, J., … Lyden, D. (2012). Melanoma exosomes educate bone marrow progenitor cells toward a pro-metastatic phenotype through MET. Nature medicine, 18(6), 883-891.
75. Qu, L., Ding, J., Chen, C., Wu, Z. J., Liu, B., Gao, Y., Chen, W., Liu, F., Sun, W., Li, X., Wang, X., Wang, Y., Xu, Z. Y., Li, Y., Fang, Z. Y., Xu, Z. D., & Bao, Y. (2016). Exosome-Transmitted lncARSR promotes sunitinib resistance in renal cancer by acting as a competing endogenous RNA. Cancer cell, 29(5), 653-668.
76. Reclusa, P., Taverna, S., Pucci, M., & Durendez, E. G. (2018). Exosome-based therapeutics in cancer: current status and future directions. Journal of personalized medicine, 8(1), 1-22.
77. Ren, J., Ding, L., Zhang, D., Shi, G., Xu, Q., Shen, S., Wang, Y., & Wang, T. (2019). Carcinoma-associated fibroblast-derived exosomes regulate tumor progression of hepatocellular carcinoma via miR-200a-3p/Sec23a signaling. Journal of hematology & oncology, 12(1), 1-14.
78. Ridder, K., Keller, S., Dams, M., Rupp, A. K., Schlaudraff, J., Del Turco, D., Starmann, J., Macas, J., Karpova, D., Devraj, K., Depboylu, C., Landfried, B., Arnold, B., Plate, K. H., Höglinger, G. U., Sültmann, H., Altevogt, P., & Momma, S. (2014). Extracellular vesicle-mediated transfer of genetic information between the hematopoietic system and the brain in response to inflammation. PLoS biology, 12(6), e1001874.
79. Sauter, E. R. (2013). Exosome and microvesicle–mediated cancer detection and monitoring: prospects and limitations. Diagnostic cytopathology, 41(12), 1065-1070.
80. Shao, Y., Chen, T., Zheng, X., Yang, S., Xu, K., Chen, X., & Xu, F. (2018). Colorectal cancer-derived small extracellular vesicles establish an inflammatory premetastatic niche in liver metastasis. Carcinogenesis, 39(11), 1368-1379.

81. Simpson, R. J., & Mathivanan, S. (2012). Extracellular microvesicles: the need for internationally recognised nomenclature and stringent purification criteria. Journal of proteomics and bioinformatics, 5(4), i-ii.
82. Skotland, T., & Sandvig, K. (2019). Transport of nanoparticles across the endothelial cell barrier. Nano today, 25, 85-96.
83. Soung, Y. H., Ford, S., Zhang, V., Chung, J., & Exosomes in Cancer Diagnostics. (2017). Current translational geriatrics and experimental gerontology reports, 6(2), 89-94.
84. Syn, N. L., Wang, L., Chow, E. K., Lim, C. T., & Goh, B. C. (2016). Exosomes in cancer nanomedicine and immunotherapy: prospects and challenges. Trends in biotechnology, 34(12), 983-995.
85. Théry, C., Witwer, K. W., Aikawa, E., Alcaraz, M. J., Anderson, J. D., Andriantsitohaina, R., Antoniou, A., Arab, T., Archer, F., Atkin-Smith, G. K., Ayre, D. C., Bach, J. M., Bachurski, D., Baharvand, H., Balaj, L., Baldacchino, S., Bauer, N. N., Baxter, A. A., … Zuba-Surma, E. K. (2018). Minimal information for studies of extracellular vesicles 2018 (MISEV2018): a position statement of the International Society for Extracellular Vesicles and update of the MISEV2014 guidelines. Journal of extracellular vesicles, 7(1), 1535750.
86. Tkach, M., & Théry, C. (2016). Communication by extracellular vesicles: where we are and where we need to go. Cell, 164(6), 1226-1232.
87. Vader, P., Mol, E. A., Pasterkamp, G., & Schiffelers, R. M. (2016). Extracellular vesicles for drug delivery. Advanced drug delivery reviews, 106, 148-156.
88. Vagner, T., Spinelli, C., Minciacchi, V. R., Balaj, L., Zandian, M., Conley, A., Zijlstra, A., Freeman, M. R., Demichelis, F., & De Carvalho, D. D. (2018). Large extracellular vesicles carry most of the tumour DNA circulating in prostate cancer patient plasma. Journal of extracellular vesicles, 7(1), 1505403.
89. Wang, C., & Wang, M. (2019). Exosomes released by hepatocarcinoma cells endow adipocytes with tumor-promoting properties. Journal of hematology & oncology, 12(1), 1-12.
90. Wu, Y., Deng, W., McGinley, E. C., Klinke, D. J., & Melanoma exosomes deliver a complex biological payload that upregulates PTPN11 to suppress T lymphocyte function. (2019). Pigment cell & melanoma research, 32(5), 627-638